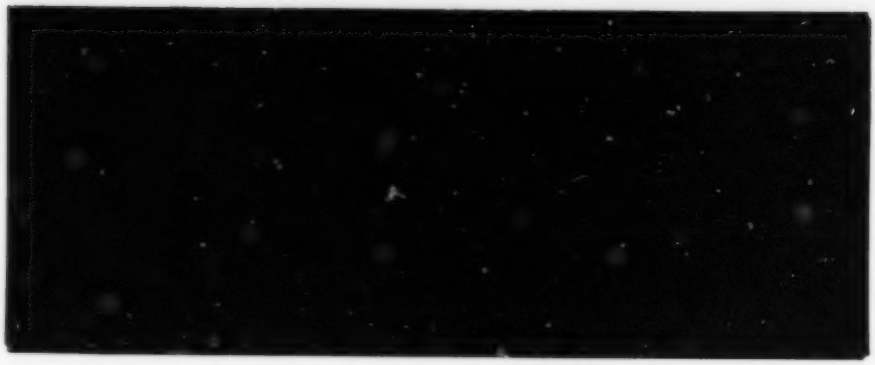
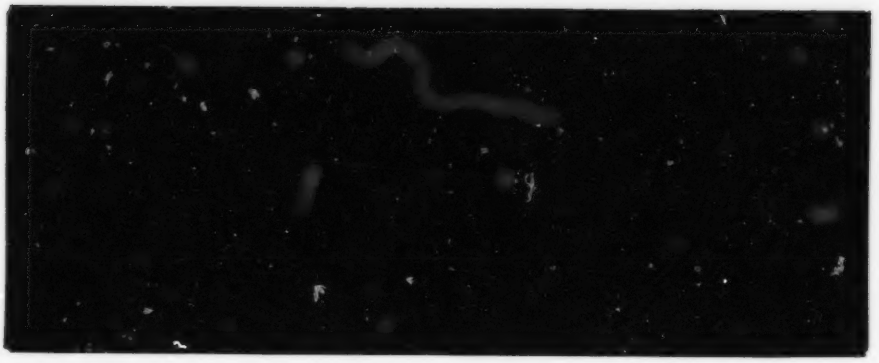
Saskatchewan
Agriculture

Agriculture Development Fund

AOFS0043





91-02628/

Technical/Final Report
to
Saskatchewan Agriculture Research Fund

The Roles of Insulin, Glucose and Free Fatty Acids in the Development of Ketosis and Fatty Liver Disease in Ruminants

S-85-01-0019/5-85-62

FINAL REPORT

5-8501-0019

Project No. 5-85-62

The Roles of Insulin, Glucose and Free Fatty Acids in the Development of Ketosis and Fatty Liver Disease in Ruminants

The control of body fat mobilization in dairy cattle is a complex process which often results in ketosis and/or fatty liver disorders. These disease states occur when an animal cannot obtain sufficient energy from the diet to meet the energy requirements of pregnancy or heavy lactation. Normally any energy deficit is supplied primarily by mobilization of body fat. The control mechanisms regulating the rate of free fatty acid (FFA) release from fat reserves are poorly understood. It appears however, that an excessively high rate of release is a major factor in the development of these disease states.

The initial objective of the proposed research was to develop a FFA infusion technique which would elevate plasma FFA in sheep to levels of 1.0 to 1.5 mM. These levels are often found in ketotic animals whereas healthy individuals seldom have blood levels exceeding 0.5 mM.

One technique was developed for use in anaesthetized rats. The infusate was a milk-like emulsion formed by combining FFA with an albumin solution or serum. This type of infusate containing 3% to 5% FFA was extensively tested in our laboratory during the winter of 1985-86. After 30 individual infusions in four different animals the technique was abandoned. These infusions were deemed unacceptable due to a concurrent induction of fever in each animal. In nearly every case within 30 minutes of beginning the infusion, the animal would exhibit a mild shaking reaction followed closely by an increase in body temperature.

The second published FFA infusion technique involved the use of a continuous flow centrifuge. Blood was continuously drawn from the animal (dog)

into a centrifuge which separated cells and plasma. FFA were then infused into the plasma line. The FFA enriched plasma (0.2% FFA) was then recombined with the cells and continually infused into the animal at rates up to 100 ml/min. The low concentration of FFA did not result in hemolysis and a fever response was not reported. Our laboratory is not equipped nor is the college with such a centrifuge yet we did attempt to infuse plasma enriched with low concentrations (0.5%-1.0%) of FFA at relatively high rates (5-10 ml/min).

The final infusate we developed was clear, stable and could be sterilized by filtration through a 0.22 μ filter. From May to August, 1987 a total of 52 separate infusions in nine animals were performed. A number of variables in both the infusate preparation and delivery system were investigated in an attempt to eliminate the concurrent fever response and hemolysis which usually accompanied each infusion. Unfortunately we were not successful. The effects of the infusate on plasma levels of FFA were determined after each infusion and are summarized in Table 1. At delivery rates of up to 60 mg/min FFA plasma FFA levels elevated only slightly. Even at this delivery rate hemolysis and fever reactions were encountered. Lowering the infusion rates generally avoided the hemolysis but the development of fever persisted. In addition the lower rates of course failed to elevate plasma FFA to sufficient levels.

We have thus had to abandon this approach in our study of ketosis and fatty liver diseases. Although numerous alternatives were attempted in our efforts to develop a model to study this metabolic problem, the infusion of exogenous FFA to elevate blood levels is impractical. It is also our conclusion that the reports in the literature are misleading and fail to account for the affects of fever and hemolysis which nearly always occur with these infusions.

Table 1. The effects of intravenous infusions of FFA at various rates on the plasma concentrations of FFA in sheep. Data are from 12 representative infusions in which plasma FFA levels were determined.

Date	Infusion			Maximum Elevation of Plasma FFS (mM)	Side Effect
	FFA Concentration (%)	rate (mls/min)	rate (mg/min)		1 - fever 2 - hemolysis
29/4/86	1.0	4.0	40	0.10	1
1/5/86	1.0	4.0	40	0.07	1
2/5/86	1.8	3.0	54	0.07	1
8/5/86	0.8	5.0	40	0.06	1
21/5/86	0.8	5.0	40	0.03	1
4/7/86	1.0	5.0	50	0.05	1,2
8/7/86	1.0	5.0	50	0.05	1,2
15/7/86	0.75	11.2	84	0.18	1,2
16/7/86	0.75	8.4	63	0.12	1,2
23/7/86	0.75	7.6	57	0.07	1,2
29/7/86	0.75	8.5	64	0.14	1,2
7/8/86	0.90	8.4	75	0.12	1,2